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Response of benthic cave invertebrates to organic pollution events

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Abstract

1. Even though the fragility and vulnerability of subterranean ecosystems (caves, groundwater and hyporheic habitats) is widely acknowledged, the impacts of anthropogenic disturbances have been poorly quantified when compared to surface waters. In particular, limited data exist regarding the impact of organic pollution upon aquatic cave invertebrate communities.

2. The Peak-Speedwell Cavern system (Derbyshire, UK) was affected by two organic pollution events, during a 7-year study (1997-2003), originating from the same source in the surface catchment but resulting in markedly different ecological responses. The first event led to the elimination of most taxa from affected sites while the second resulted in an increase in abundance of organisms within the cave associated with the increased availability of trophic resources. The second event also coincided with the invasion of the stygophilic amphipod, *Gammarus pulex*, at a site where it had not previously been recorded.

3. Recovery of the invertebrate community following both organic pollution events occurred within 12-months. Recolonisation of the affected sites was facilitated by annual flooding of the cave and by the presence of refugia on unaffected subterranean tributaries.

4. The data highlight the problems associated with the conservation and management of subterranean ecosystems where impacts in distant surface catchments may have unseen repercussions for the subterranean environment. Aquatic subterranean habitats are not widely monitored and the impacts of pollution/disturbance may not be detected in surface waters for some time, if at all, due to dilution effects. Caves supporting obligate subterranean organisms (stygobites) are particularly vulnerable to these pressures and require clear management strategies to protect both the subterranean and surface catchments which support them.
Introduction

The dark zones of caves are naturally typified by low organismal abundance and diversity (Holsinger, 1988; Jasinska et al., 1996; Culver and Sket, 2000), and due to the relatively constant abiotic conditions, their biological communities are widely considered to be more stable compared with those in epigean systems (Culver, 1985; Simon et al., 2003). In the absence of light and primary producers, cave habitats are largely oligotrophic, relying almost exclusively on dissolved or particulate organic matter originating in surface (epigean) habitats (Poulson and Lavoie, 2000; Simon et al., 2003). Hence, it is highly likely that changes in landuse and/or management practices within surface epigean catchments may result in significant changes to the trophic dynamics of subterranean (hypogean) food webs (Poulson and Lavoie, 2000; Hancock et al., 2005). Consequently, aquatic subterranean habitats (hyporheic zone, groundwater and wet caves) are considered to be vulnerable to anthropogenic activities (Sket, 1999; Gunn et al., 2000; van Beynen and Townsend, 2005; Boulton, 2005), yet our understanding of the impacts of such activities upon subterranean ecosystems is much more limited compared with epigean waterbodies (Elliott, 2000; Hancock et al., 2005).

Although there has been some recent increase in interest regarding the impact of disturbances upon biological communities within groundwater aquifers (e.g., Danielopol et al., 2003; Hancock, 2002), research exploring the influence of disturbances upon cave ecosystems has been limited despite wide recognition of their high conservation/biodiversity value (Culver and Sket, 2000). Anthropogenic disturbances and modifications of cave ecosystems associated with heavy metals (Graening and Brown, 2003), faecal bacteria (Green et al., 1990; Simon and Buikema, 1997; Graening and Brown, 2003) and waste disposal (Halliday, 2003)
have been reported. However, the response of cave communities and individual species to organic pollution remains poorly quantified. This paucity of information reflects the absence of pre-disturbance baseline data and/or absence of adjacent control sites which could be used to determine the nature and magnitude of impacts. Those studies that have documented the response of cave invertebrate communities and individual species to organic enrichment and pollution show that responses are variable (Table 1). Significant changes to the structure of cave benthic invertebrate communities, particularly reductions in abundance or exclusion of obligate subterranean aquatic fauna (stygobites) as a result of organic pollution have been reported (Culver et al., 1992, Simon and Builkema, 1997, Graening and Brown, 2003). However, in some instances there have been increases in the abundance of obligate subterranean (hypogean/stygobitic) fauna and/or an increase in species richness of epigean/stygophilic faunal populations within caves, particularly when trophic resource availability is enhanced (Holsinger, 1966, Sket, 1977, Simon and Builkema, 1997, Graening and Brown, 2003). It has even been suggested that mild organic enrichment may be beneficial to stygobitic populations under some circumstances, provided that highly competitive stygophiles, epigean taxa able to complete their life-cycles within the cave but usually occurring in surface waters, do not invade (Sket, 1999, Graening and Brown, 2003).

Pollution of groundwater dominated habitats has been implicated as one of the greatest threats to the long term provision of groundwater resources and subterranean biodiversity (e.g., Boulton, 2005; Danielopol et al., 2003; Hancock et al., 2005) and in particular, cave ecology (e.g., Gunn et al., 2000; Finlay et al., 2006; Panno et al., 2006). However, data clearly demonstrating the ecological impact of pollution within caves are limited due to the difficulties associated with conducting research within subterranean habitats, the absence of pre-disturbance (pollution) data and/or information regarding the source and nature of pollutants (Gunn et al., 2000). Here we examine the response of freshwater cave invertebrates
to two point-source organic pollution events that occurred during a seven-year study (1997-
2003). Our main aims were to gauge the impact of pollution by: (1) quantifying the
invertebrate community response to pollution episodes and comparing the impacts of separate
events; and (2) investigating changes to the local populations (i.e. extinctions or invasions)
resulting from pollution.

Methods

Study Site

The study was undertaken from 1997-2003 within the Peak-Speedwell Cave system,
Derbyshire (UK). Peak Cavern and Speedwell Cavern are interconnected and contain more
than 16 km of active (wet) and relict (dry) cave passages that have formed within
Carboniferous limestone (karst geology). There is limited hydrological connectivity between
the caves, except under high flow (flood) conditions, when water from Speedwell Cavern may
rise into the higher passages within Peak Cavern. Water within Peak Cavern is largely derived
from autogenic sources (water that has only been in contact with limestone bedrock and
overlying soil, and percolates into the cave) which are concentrated into two main
subterranean streams that enter the cave from flooded conduits, Ink Sump and Far Sump.
These streams flow along the Peak Cavern streamway, enter another flooded conduit and
emerge as Peak Cavern Rising, a large spring at the head of Peakshole Water (Figure 1).
Water within Speedwell Cavern is largely derived from allogenic sources - twelve streams
that flow on the surface over non-limestone geologies before sinking underground. The
streams combine underground, enter Speedwell Cavern via two flooded conduits, Main
Rising and Whirlpool Rising, flow through the cave, enter another flooded conduit and
finally emerge from two springs, Russet Well and Slop Moll, which both flow into Peakshole
Water (Gunn et al., 2000). Landuse in both catchments is dominated by livestock grazing,
which has historically resulted in inputs of faecal bacteria to the subterranean ecosystem (Gunn et al., 1998, Hunter et al., 1999).

Detection of pollution and tracing the source

Two major point source pollution episodes were experienced during the study period: (1) during early 1999; and (2) between December 2001-January 2002. Both events occurred when parts of the cave were inaccessible due to flooding and as a result the passage of the pollutant could not be directly monitored in situ. Following the detection of pollution within Peak Cavern due to the first event a survey of the surface catchment identified an orange liquor draining from a large mound into a small stream-sink. The pollutant was organic rich material which was being stock-piled prior to spreading on land as an ameliorant and was principally composed of paper pulp and organic rich peat from a water treatment works. This material formed a mound that covered >500 m² to a depth of at least 1m. When the pollution became evident within Peak Cavern the landowner was asked to take action to prevent runoff/pollution entering the cave. However, the second pollution event occurred after this same material had been partially dispersed on the surface catchment. Once the soils were in a saturated state, following heavy rain in late 2001, water re-entered the same sink holes leading to further degradation.

The cave passage downstream of Ink Sump (Figure 1) was heavily stained following both events and the substratum was covered by an orange residue. The staining was observed and reported by recreational cavers and divers but no visible evidence of pollution was detected outside the cave within the springs or river draining the caves (Peakshole Water) during the first event. The second pollution event occurred over a longer period but discolouration of the water was only observed for 24 hours. Hydrological connectivity between the pollutant and the cave was demonstrated by a tracing experiment using two fluorescent dyes, sodium...
fluorescein (CI 45359 Acid Yellow 73) and rhodamine WT (CI Acid Red 388). The dyes were detected at the head of Ink Sump, the most upstream visible point where pollution was recorded within Peak Cavern and also entered Far Sump (the second major percolation input to Peak Cavern - Figure 1). The experiment also indicated that a large proportion of the tracer (and therefore the pollutant) travelled ~4 km in an easterly direction and was discharged by a natural spring and two anthropogenic sources (soughs) draining water from disused lead mines, and that a small volume of tracer also entered Speedwell Cavern (Wood et al., 2002).

Microfloral analysis of water samples from Peak Cavern indicated the presence of a number of cellulose degrading bacteria associated with the biodegradation of the paper pulp following the second event (Hibberd, 2003).

**Monitoring and laboratory processing**

The invertebrate community was routinely sampled monthly over the 7-year period (84 months; January 1997- December 2003) from 5 sites within Peak Cavern and from the Peak Cavern Rising (n = 480) and from 6 sites (n = 472) within Speedwell Cavern (Figure 1). Benthic invertebrates were sampled using a 0.05 m² cylinder sampler (fitted with a 90 μm mesh net) over a 30-second period. Additional examination of larger clasts within the cylinder was also undertaken, where they occurred. Due to the potential disturbance and degradation associated with extensive sampling of subterranean habitats single cylinder samples were collected and sampling occasions were used as replicates (Gunn et al., 2000). Sampling could not be undertaken at all sites each month due to flooding of some subterranean passages during the winter and early spring months (5 months within the 84-month study period). At Peak Cavern, three sites were all downstream of the pollution source (Figure 1 – Peak Polluted: PP1, PP2 and PP3) and three sites (control sites) were located on unaffected tributaries (Peak Control: PC1, PC2 and PC3).
All specimens were preserved in the field with 70% industrial methylated spirits (IMS) and returned to the laboratory for processing and identification. Samples were washed and screened on 250μm and 90μm mesh sieves. Material >250μm was manually inspected by removing all invertebrates from an illuminated sorting tray. All sediment retained on the 90μm mesh sieve was examined in a grooved (5 mm) Bogorov sorting tray at 10-50 magnifications to ensure all material from the samples was examined. All macroinvertebrate taxa were identified to species level where possible. Chironomidae, Oligochaeta and Copepoda specimens were examined individually and mounted on microscope slides for examination (up to 400 magnifications) as required for species level identification.

Water temperature (°C), conductivity (μS cm⁻¹), pH and dissolved oxygen (mg l⁻¹) were measured in the field using a portable YSI 600R water quality probe. Replicate water samples were collected from the caves and associated springs and analysed for nitrate (mg l⁻¹) and phosphate (mg l⁻¹) concentrations. Preliminary analysis indicated that there were no significant differences between samples pre- and post-pollution, or between those sites affected by the pollution and those on unaffected tributaries. This reflects the fact that the pollution entered the cave on the flood hydrograph when most of the cave was inaccessible.

Data analysis
Differences in the invertebrate community between the two caves, and sites affected and unaffected by pollution were examined on an annual basis (calendar year January-December). This corresponded to the timing of flood events and the detection of pollution (disturbance events) within Peak Cavern, and provided 2-years of pre-disturbance data (1997 and 1998), 2-years when pollution events occurred (1999 and 2002), and 3 other years (2000, 2001 and 2003). The invertebrate community was characterised by the following metrics: total abundance (individuals m⁻²), number of taxa, Shannon-Wiener diversity index and the Berger
Parker dominance index. The latter two indices were calculated using the α Species Diversity and Richness software (Pisces Conservation, 1998). Preliminary examination of the data for the different sites and years using Levene’s test for homogeneity of variances were significant for some groups (P<0.05). Hence, the non-parametric Kruskal-Wallis test was applied to examine differences between the caves, polluted and control sites, and for the different time periods.

Results

Invertebrate community

A total of 34 aquatic invertebrate taxa were recorded during the study period (Table 2). The pre-disturbance Peak Cavern invertebrate community was dominated by Oligochaeta (5 taxa: *Limnodrilus hoffmeisteri*, *Lumbriculus variegatus*, *Spirosperma ferox*, *Stylodrilus* sp. and *Tubifex tubifex*) and Copepoda (4 taxa: *Acanthocyclops venustus*, *A. vernalis*, *Diacyclops bicuspidatus* and *Megacyclops viridis*) in terms of abundance. Other invertebrate taxa typically comprised less than 15% of the total abundance for individual sampling occasions. The community within Speedwell Cavern was more variable and was dominated by Oligochaeta (*Limnodrilus hoffmeisteri* and *Tubifex tubifex*), Chironomidae (particularly two Orthocladiinae: *Rheocricotopus fuscipes* and *Brillia modesta*), Copepoda (*Acanthocyclops venustus*, *A. vernalis*, *Diacyclops bicuspidatus*) and the amphipod *Gammarus pulex*. Faunal abundance displayed seasonal variability, demonstrating the influence of epigean inputs of water and organic matter. Examination of the invertebrate communities for the pre-disturbance period (1997 and 1998) indicated that samples from Speedwell Cavern supported a greater abundance of invertebrates, although the community was dominated by a smaller number of taxa compared to Peak Cavern (Kruskal-Wallis test: abundance - P<0.001, Berger Parker dominance – P<0.005). Samples from Peak Cavern supported a greater number of taxa and had a higher Shannon-Wiener diversity than Speedwell Cavern (Kruskal-Wallis test:...)
number of taxa – P<0.001; Shannon-Wiener - P<0.001 – see Wood et al., 2002 for further
details).

Pollution episode 1

At sites affected by pollution in Peak Cavern no benthic invertebrates were recorded in the
month after the pollution event, although a large number of dead and decaying earthworms
(Lumbricus terrestris) were recorded at the channel margins. The abundance of freshwater
taxa remained low at polluted sites for the rest of the year compared with control sites (Figure
2), with the invertebrate community being almost exclusively composed of two oligochaetes
(Limnodrilus hoffmeisteri and Tubifex tubifex). The first pollution episode resulted in a
significant reduction in the abundance at affected sites (1999 in Figure 3a) compared to pre-
disturbance data (1997 and 1998 in Figure 3a) and control sites (Figure 3b). A similar pattern
was observed for the number of taxa and the Shannon-Wiener diversity index, and an inverse
pattern for the Berger-Parker dominance index (see Table 3 for pair-wise comparisons). No
significant differences in benthic abundance, number of taxa, Shannon-Wiener diversity or
Berger Parker dominance were recorded between the polluted and control sites within Peak
Cavern in the following 2 years (2000 and 2001) (Table 3), and there were no differences in
any invertebrate community parameters for Speedwell Cavern between the pre-disturbance

Pollution episode 2

As a result of the second input of pollutant in 2002, a significant increase in benthic
abundance occurred at the affected sites within Peak Cavern (Figure 3a) compared with two
of the control sites (Figure 3b). A similar pattern was observed for the Berger-Parker
dominance index and an inverse pattern for the number of taxa and the Shannon-Wiener
diversity index (see Table 3 for pair-wise comparisons). The abundances of two oligochaetes
(Limnodrilus hoffmeisteri and Tubifex tubifex) increased significantly (to >500 m⁻²) within one month of the input of the pollutant (Kruskal-Wallis test – P<0.001). In the following months, numbers of the epigean amphipod, Gammarus pulex, also increased significantly at polluted sites compared with two control sites (Kruskal-Wallis test – P<0.001). In February 2002, G. pulex was recorded for the first time during the study period at one control site (PC1 in Figure 1). Following the discovery of G. pulex at the site the total abundance of invertebrates, particularly Oligochaeta and Copepda, was reduced compared to the other control sites (Figure 4a-e). No significant differences between any invertebrate community parameters were recorded for Speedwell Cavern following the second pollution episode.

Discussion

The nature of cave pollution and disturbance

Differences in the physical nature of perturbations, in the form of pulse, press and ramp disturbances can result in multiple and markedly different biotic responses within aquatic ecosystems (sensu Lake, 2000). Pollution disturbances of groundwater dominated ecosystems can be associated with both press and pulse disturbances. Press disturbances are typically associated with the diffuse entry of material from a relatively large geographical area which percolates into the subterranean groundwater environment (Hancock et al., 2005; Rinaudo et al., 2005). Pulse events are usually associated with the rapid transfer of material into the subterranean environment from a specific location within the surface catchment and may be associated with high water input (Culver et al., 1992; Graening and Brown, 2003). Both of the events recorded in this investigation were clearly point-source disturbances associated with flood events. However, flood events occurring between the two pollution events, during 2000 and 2001, did not appear to result in any significant input of pollutant and acted as ‘flushing flows’ which facilitated the recovery of the benthic invertebrate community (abundance, number of taxa, diversity and dominance) to pre-disturbance levels (Figure 3).
Both pollution events resulted in significant changes to the benthic invertebrate community of affected sites within Peak Cavern. However, no impact was recorded within the adjacent system (Speedwell Cavern) despite water tracing experiments indicating limited hydrological connectivity with the stream-sink through which the pollutant entered the groundwater system (Wood et al., 2002). This reflects the different hydrological characteristics of the two caves. Water in Speedwell Cavern is primarily derived from sinking streams and as a result the residence time of water within the cave is short, dissolved and particulate organic matter input is relatively high, and pollutants are likely to be diluted and transported through the system relatively quickly (Gunn et al., 2000; Simon et al., 2003). In contrast, water within Peak Cavern is principally derived from percolation water that has passed through the overlying soil and rock and, as a result, the residence time of water is longer. In addition, the volume and delivery of dissolved and particulate organic matter and abundance of invertebrates is usually lower within percolation water dominated systems such as Peak Cavern (Poulson and Lavoie, 2000; Simon et al., 2003). These natural hydrological characteristics reflect a well know gradient of differences that strongly influences the volume, timing and processing rate of trophic resources within subterranean ecosystems (e.g., Poulson and Lavoie, 2000; Simon and Benfield, 2001, Simon et al., 2003).

Faunal response to pollution

Faunal response to the pollution events was marked and indicative of significant disturbance events. Direct faunal community response to the pollution of caves has only been recorded in a limited number of previous studies (e.g. Culver at al., 1992), with several studies comparing degraded systems with reference sites in the absence of non-affected control sites (e.g. Holsinger, 1966, Simon and Buikema, 1997). Few studies have included detailed pre- and post-disturbance data or have been undertaken over a comparable length of time. The greatest
changes to the invertebrate community of Peak Cavern were associated with a limited number of taxa (2 oligochaetes: *Limnodrilus hoffmeisteri* and *Tubifex tubifex*, the amphipod *Gammarus pulex* and 4 Copepoda - *Acanthocyclops venustus*, *A. vernalis*, *Diacyclops bicuspidatus* and *Megacyclops viridis*).

During the period immediately following both pollution episodes the invertebrate community at affected sites was dominated by the oligochaetes *Limnodrilus hoffmeisteri* and *Tubifex tubifex*. Both of these taxa are widespread, occur in most surface waters and have been recorded from caves across the globe where they have been associated with organic enrichment (Swayne et al., 2004; Wetzel and Taylor, 2001). During the first pollution event densities were lower than baseline conditions (<50 m$^{-2}$) and during the second event they were significantly higher (>200 m$^2$) at degraded sites (Figure 3). Their dominance of the invertebrate community within Peak Cavern during these events suggests they are relatively resilient and good indicators of organic pollution within caves and other groundwater dominated ecosystems (Lafont et al., 1996; Lafont and Vivier, 2006).

The increased abundance of cyclopoid copepods following the input of organic material during the second event probably reflects an increased food supply for these taxa. Several cyclopoid copepods (including those in the genus *Acanthocyclops*) are known to be predatory (Fryer, 1957; Galassi et al., 2002), feeding on taxa such as ciliates, rotifers, small oligochaetes and other small crustaceans, all of which may have increased abundances in conditions of high organic matter. At the same time, other cyclopoid taxa are more reliant on fine detrital material (Galassi et al., 2002) that, again, is likely to be more plentiful during an organic pollution event. The abundance of *Gammarus pulex* also increased (>20 individuals m$^{-2}$) at affected sites as a result of the second pollution episode, as well as invading one of the adjacent control sites. *G. pulex* have been recorded in many cave systems in the UK, where
they frequently occur in relatively high abundances (Proudlove et al., 2003). Stygophilic
gammarids have been recorded within a number of caves around the world where some
populations display adaptations to the subterranean environment (e.g. Culver et al., 1995).
Epigean *Gammarus* species have been widely reported to be highly competitive and invasive
in some instances (MacNeil et al., 2003). It is now widely acknowledged that some
gammarids are omnivorous and may be active and effective predators (Kelly et al., 2002) and
the invasion of epigean (stygophilic) taxa into subterranean habitats may result in the
displacement and/or elimination of hypogean (stygobitic) taxa (Skeet, 1977).

The aquatic invertebrate communities of both caves were almost exclusively composed of
stygophiles, and none can be regarded as obligate subterranean taxa (stygobites); although the
larvae of the dytiscid beetle *Hydroporus ferrugineus* has only been recorded from the Peak-
Speedwell system and may be an obligatory subterranean life stage (Alarie et al., 2001). No
stygobitic taxa have been recorded from 48 karstic springs within the wider limestone region
of the English Peak District (Wood et al., 2005), suggesting that the absence of stygobitic
fauna from the Peak-Speedwell Cavern system is not due to pollution alone. Absence of
hypogean taxa may reflect glacial activity during the Pleistocene, the maximum extent of
which was thought to mark the limits of subterranean faunal distributions. However, there is
increasing evidence that stygobitic fauna persisted in sub-glacial refugia beneath the ice in
many areas (e.g. Holsinger et al., 1997) including the UK where stygobitic taxa have been
recorded some distance north of the maximum extent of glaciation (Proudlove et al., 2003;
Bratton, 2006).

*Parallels and contrasts between pollution events*

A number of parallels and contrasts between the events and their impact on the cave
ecosystem can be identified. Both of the pollution events recorded during the study period

coincided with floods and originated from the same location within the surface catchment. The impact on the benthic community at polluted sites was rapid (one month following their detection) and persisted until the next major flood event. However, the response of the community and individual taxa to the two events was markedly different. The first pollution episode resulted in a significant reduction in community abundance, number of taxa and Shannon- diversity index but an increase in the Berger-Parker dominance index at affected sites. The second episodes led to a marked increase in the community abundance and Berger-Parker dominance index, and a reduction in the number of taxa, and Shannon- diversity index. The differences in the community response to the events probably reflects differences in the magnitude of the flood events and associated pollutant loading. The first event resulted in the input of pollutants which were largely contained within the cave and led to the exclusion of almost all fauna from affected sites. The second event was associated with a period of sustained high flow and it is likely that a large proportion of the pollutant was transported through the cave, and was observed as discolouration of the water emerging from Peak Cavern Rising. The pollution load retained within the cave associated with the second event was probably lower, did not lead to sub-lethal concentrations and may have actually enhanced the trophic resources available within the cave leading to the marked increase in the abundance of some members of the invertebrate community (Graening and Brown, 2003; Simon and Buikema, 1997; Sket, 1999). Recovery of the benthic community was relatively rapid following both pollution events, possibly due to the presence of a large number of refugia within non-polluted sites. Subsequent flooding of the cave in the proceeding years (2000 and 2003 respectively) appeared to “cleanse” the system of the pollutant and facilitated the recovery of fauna at all sites following the first event and all but one site (which was invaded by Gammarus pulex) following the second (Figure 3 and Figure 4). In other studies, recovery of aquatic cave
invertebrate communities following pollution disturbances has not been as rapid as reported in the current investigation and the impacts have persisted for some time (in excess of 3-years - see Culver et al., 1992). However, data on recovery times for cave communities are usually absent (Graening and Brown, 2003, Simon and Buikema, 1997), reflecting the long-term and diffuse nature of the impact of pollution on some systems but also the fragility of cave ecosystems, the difficulty of undertaking research in subterranean environments and the paucity of pre-disturbance baseline data available for most systems. In the case of the Peak-Speedwell Cavern system, the relatively rapid recovery may have reflected the legacy of impacts upon the subterranean ecosystem (Gunn et al., 2000). This may also explain the absence of stygobitic taxa which are less competitive and more vulnerable to pollution disturbances than most stygophilic taxa (Graening and Brown, 2003; Panno et al., 2006; Sket, 1999).

Implications for conservation and management

Managing groundwater/subterranean ecosystems is particularly difficult since the most damaging activities usually occur in the surface catchment (van Beynen and Townsend, 2005; Danielopol et al., 2003; Gunn et al., 2000). There may be an extended time-period between a disturbance event occurring in the surface catchment and its detection within the subterranean system, by which time irreversible damage may have already occurred (Hancock et al., 2005). Even after the detection of any pollutant, tracing the source may be problematic because the pollution may have ceased and/or the input may be episodic, as recorded in the current investigation.

In Great Britain (England, Wales, Scotland), the major mechanism for legally protecting, and thereby conserving wildlife and earth science features is through notification as a ‘Site of Special Scientific Interest’ (SSSI). A list of “operations likely to damage the special interest”
is issued to each owner of land in the boundaries of a SSSI at the time the site is designated
and the relevant country authority (Natural England, Countryside Council for Wales, Scottish
Natural Heritage) must be consulted before any of the listed operations are undertaken. If it is
considered that the proposed action will damage the scientific interest of the site then
permission may be denied and the authority may enter into a management agreement with the
land owner. Following a Geological Conservation Review (GCR) which began in 1977 (Ellis
et al., 1996) 48 ‘cave’ sites were identified and subsequently have been designated as SSSI.
Descriptions and evaluations of the geomorphological evolution of each Cave and Karst GCR
site have been published (Waltham et al., 1997). At the time of the GCR the boundaries of the
48 sites encompassed 879 named caves, ~30% of the total caves in Britain (Hardwick and Gunn,
1996). These 879 caves included all of the longer cave systems so that ~75% of known cave
passage (and hence of the total cave resource) was within areas proposed for conservation.

Some of the caves designated as SSSIs in Great Britain were based on their biological interest,
although almost exclusively on the basis of bats (Chiroptera) and/or bat roosts. Aquatic
invertebrates are only listed as an additional reason for notification at one site (Pridhamsleigh
Cave SSSI, Devon) where Niphargus glenniei (Crustacea: Amphipoda), an endemic amphipod
which is abundant within the cave, occurs. The Peak-Speedwell Cavern system forms part of a
Site of Special Scientific Interest (SSSI) but its designation only covers the earth science
interests and does not include any subterranean ecological/biological interests (Gunn et al.,
2000). However, designation of a cave SSSI does provide limited, even if unintentional,
protection for aquatic cave ecosystems and the communities they support because each SSSI has
a list of ‘operations requiring consent’. These have been drawn up to protect the earth science
features of interest but by providing controls on water quality and water quantity they may also
benefit the whole subterranean ecosystem. Initially the protection of sites was confined to
operations on the overlying land surface and the land owner was held responsible for any
infringement. However, in England and Wales, part of the Countryside and Rights of Way Act 2000 (CROW) makes it possible for action to be taken against any person damaging the scientific interest of a SSSI even if the action took place outside the SSSI boundaries. As the current research demonstrates, this is particularly important in the case of active cave systems that often receive inputs of water from surface streams whose catchment is outside of the SSSI. All notified water pollution incidents, whether outside or inside of a SSSI, are subject to investigation by the Environment Agency (England and Wales) or the Scottish Environment Protection Agency. However, if the investigating agency is unaware of the composition, sensitivity or even existence of potentially vulnerable aquatic communities in caves then their conservation is not likely to be considered. Knowledge regarding subterranean biodiversity and its conservation value in the UK is severely limited due to an absence of historic and contemporary scientific research compared to other geographical localities (e.g., Ferreira et al., 2007; Culver et al., 2000) and therefore requires an urgent reassessment.

Many obligate aquatic subterranean organisms (stygobites) are confined to relatively small geographical locations (Christman et al., 2005; Ferreira et al., 2007), and display morphological and physiological adaptations to their environment (Coineau, 2000; Culver et al., 1995). As a result, many aquatic cave communities are scientifically important and of high conservation value (Sket, 1999). Managing and mitigating the effects of organic pollution within groundwater dominated habitats may be particularly difficult due to the highly diffuse nature in which many pollutants enter aquifers and cave ecosystems (Boulton, 2005; Sket, 1999) and the long residence time of water compared to epigean riverine systems. Across North America and in some Europe countries, a greater awareness of subterranean biodiversity exists (Culver et al., 2000; Ferreira et al., 2007; Sket, 1999), and some faunal species have been recognised as threatened by the International Union for Conservation of Nature and Natural Resources (IUCN 2006). However, conservation of subterranean fauna is
problematic since while individual species and caves may be protected, the wider community and the surface catchment usually have limited or no protection.

There is a growing need to consider the importance of groundwater quality within subterranean systems since it has major implications for obligate subterranean taxa, and may ultimately have a significant impact on surface waters and their ecology (Boulton, 2005, Hancock, 2002). However, the identification of indicator organisms and the development of biotic indices for groundwater dominated ecosystems, including caves, are currently limited (e.g. Lafont et al., 1996; van Beynen and Townsend, 2005; Hahn, 2006). Greater awareness regarding the impact and implication of disturbances, particularly pollution, upon groundwater dominated ecosystems is required. Given the limited biological monitoring of subterranean groundwater dependant ecosystems, and the largely unseen consequences of pollution within them, a significant knowledge gap exists regarding their impacts. Future research should address these issues to ensure the continued conservation and protection of subterranean faunal communities and the subterranean and surface water ecosystems within the wider drainage basin.

Acknowledgements

P JW acknowledges the support of the British Ecological Society (Small Ecological Project Grant – 1371) and the Natural Environment Research Council (NER/M/S/1999/00152 and GR8/04287) for funding parts of this research. Thanks to Dr Paul Hardwick, Laura Chapman, Garry Rushworth, Robin Kenyon and Richard Battye for field and laboratory assistance and to John Harrison and Tony Marsden for providing access to the caves. Thanks to Prof. P. Armitage, J. Blackburn, G. Fryer, M. Greenwood and Dr D. Horne for confirmation of faunal identifications. Thanks to Ben Le Bas (Natural England) and David Ottewell (Environment
Agency for comments on cave conservation and management; and to Phil Boon and an
anonymous reviewer for comments on a draft of this manuscript.

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Cedars, Lee County, Virginia, an ecologically significant and threatened karst area. In
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Lymington, Hampshire, UK.


List of Figures

Figure 1. The Peak-Speedwell Cavern system indicating the location of invertebrate sampling sites within Speedwell Cavern (1-6), Peak Cavern control sites (PC1-PC3), polluted sites (PP1-PP3) and other specific locations referred to within the text.

Figure 2. Mean invertebrate community abundance (individuals m\(^{-2}\) ± 1 SE) within Peak Cavern (January 1999-December 1999) for: (a) control sites and (b) polluted sites.

Figure 3. Mean invertebrate community abundance (individuals m\(^{-2}\)) and 95% confidence intervals for the Peak Cavern benthic invertebrate community (January 1997-December 2003) for: (a) polluted sites; and (b) control sites. * Indicates control site (PC1) not included in the series due to invasion of the site by *Gammarus pulex*.

Figure 4. Invertebrate community abundance for unpolluted control sites within Peak Cavern (January 1999-December 1999): (a) mean abundance of all taxa (individuals m\(^{-2}\) ± 1 SE) from control site 2 and 3 (PC2 and PC3); (b) abundance (individuals m\(^{-2}\)) of all taxa from control site 1 (PC1); (c) mean abundance of dominant Oligochaeta (*Limnodrilus hoffmeisteri* and *Tubifex tubifex* individuals m\(^{-2}\) ± 1 SE) from control site 2 and 3 (PC2 and PC3); (d) abundance of dominant Oligochaeta (*Limnodrilus hoffmeisteri* and *Tubifex tubifex* individuals m\(^{-2}\)) from control site 1 (PC1); (e) mean abundance of dominant Copepoda (*Acanthocyclops venustus*, *A. vernalis*, *Diacyclops bicuspidatus* and *Megacyclops viridis* individuals m\(^{-2}\) ± 1 SE) from control site 2 and 3 (PC2 and PC3); and (f) abundance of dominant Copepoda (*Acanthocyclops venustus*, *A. vernalis*, *Diacyclops bicuspidatus* and *Megacyclops viridis* individuals m\(^{-2}\)) from control site 1 (PC1). Solid line indicates the timing of pollution input and dashed line indicates the first record of *Gammarus pulex* at control site 1 (PC1).
1. Near Canal  
2. Pit Props  
3. Main Streamway (a)  
4. Main Streamway (b)  
5. Below Whirlpool  
6. Above Whirlpool  
C1. Mucky Ducks  
C2. Lumbago Pool  
C3. Peak Cavern Entrance  
P1. Buxton Water  
P2. Five Arches  
P3. Peak Cavern Rising  

A = Slop Moll  
B = Russet Well

Figure 1
Figure 4

(a) Abundance (individuals m$^{-2}$) vs. Month

(b) Abundance (individuals m$^{-2}$) vs. Month

(c) Abundance (individuals m$^{-2}$) vs. Month

(d) Abundance (individuals m$^{-2}$) vs. Month

(e) Abundance (individuals m$^{-2}$) vs. Month

(f) Abundance (individuals m$^{-2}$) vs. Month
Table 1. Summary of scientific papers documenting the impact of organic pollution on aquatic invertebrate communities and fauna within cave ecosystems.

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<tr>
<th>Author</th>
<th>Location</th>
<th>Pollution</th>
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<tbody>
<tr>
<td>Culver et al. 1992</td>
<td>Thompson Cedar Cave, Virginia, USA</td>
<td>Sawdust and Bark from Sawmill Operation</td>
<td>Elimination of stygobitic amphipod and isopod populations. An increase in the abundance of epigean (stygophilic) Oligochaeta (Tubificidae) and Chironomidae larvae. Limited recovery three-years after the event.</td>
</tr>
<tr>
<td>Graening and Brown 2003</td>
<td>Cave Springs Cave, Arkansas, USA</td>
<td>Septic leachate, sewage sludge and cow manure suspected</td>
<td>Elimination of stygobitic amphipods although stygobitic isopods flourished.</td>
</tr>
<tr>
<td>Holsinger 1966</td>
<td>Banners Corner Cave, Virginia, USA</td>
<td>Septic leachate (sewage)</td>
<td>An increase in the abundance of stygobitic isopod and Planaridae populations at the same time an increase in abundance of epigean (stygophilic) fauna occurred.</td>
</tr>
<tr>
<td>Panno et al. 2006</td>
<td>Illinois' sinkhole plain, Illinois, USA</td>
<td>Septic leachate (sewage)</td>
<td>Elimination of a stygobitic amphipod (Gammarus acherondytes) from one polluted system and recovery in an adjacent system.</td>
</tr>
<tr>
<td>Simon and Buikema 1997</td>
<td>Banners Corner Cave, Virginia, USA</td>
<td>Septic leachate (sewage)</td>
<td>Absence of stygobitic isopods from highly polluted pools, but common occurrence in moderately and slightly polluted waters. Exclusion of stygobitic Amphipods from any polluted waters.</td>
</tr>
<tr>
<td>Sket 1977</td>
<td>Various cave systems, Dinaric Karst, Slovenia</td>
<td>Organic enrichment</td>
<td>Podpeška jama - Increase in abundance of stygobitic fauna in the absence of epigean (stygophilic) competitors - which had no access to the site. Jama v Šahnu – elimination of all stygobitic fauna and an increase in abundance of a limited number of epigean (stygophilic) taxa - primarily Oligochaeta (Tubificidae). Postonjina-Planina cave system – Increase in abundance of epigean (stygophilic) taxa further within the cave and a corresponding decline of stygobitic taxa.</td>
</tr>
<tr>
<td>Wood et al., 2002</td>
<td>Peak Cavern, Derbyshire, UK</td>
<td>Paper pulp and peat</td>
<td>Initial exclusion of all taxa and limited recovery of epigean (stygophilic) taxa 9-months after detection of pollutant.</td>
</tr>
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</table>
Table 2. Invertebrate fauna recorded from Speedwell Cavern (1997-2003), Peak Cavern (1997-2003), and during the years when pollution occurred (1999 and 2002) within Peak Cavern for affected and control sites.

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Notes: * Indicates single specimens of stygoxene (accidental) taxa recorded within the subterranean environment; a All specimens of Lumbricus terrestris recorded in the 5 months following the detection of pollution were dead and/or
decomposing; \textsuperscript{b} Taxa recorded for the first time 9-months after the detection of the pollution within Peak Cavern.
Table 3. Kruskal-Wallis pair-wise comparison between years for invertebrate community parameters at sites within Peak Cavern affected by pollution (January 1997-December 2003): a) abundance (individuals m$^{-1}$); b) number of taxa; c) Shannon-Wiener diversity index; and d) Berger-Parker dominance index. n.b. Site invaded by *G. pulex* not included in analysis of 2002 and 2003. NS = not significant, * P <0.05, ** P<0.01 and *** P<0.001.

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